AMENDMENTS TO THE CLAIMS

Please amend the claims as noted below, without prejudice to subsequent renewal. The listing of claims below replaces all prior versions, and listings, of claims in the application.

These amendments introduce no new matter and support for the amendment is replete throughout the specification and claims as originally filed. These amendments are made without prejudice and are not to be construed as abandonment or dedication of the previously claimed subject matter, or agreement with any objection or rejection of record.

Listing of Claims:

1. (Previously presented) A composition, comprising: a cell comprising an enzyme and a caged sensor for detecting an activity of the enzyme, which caged sensor comprises:

a) one or more molecules collectively comprising:

i) a substrate for the enzyme, wherein the substrate is in a first state on which the enzyme can act, thereby converting the substrate to a second state, and

ii) a first label, wherein a first signal exhibited by the first label when the substrate is in its first state is distinguishable from a second signal exhibited by the first label when the substrate is in its second state; and,

b) one or more first caging groups associated with the one or more molecules, the first caging groups inhibiting the enzyme from acting upon the substrate.

2. (Cancelled)

3. (Previously presented) The composition of claim **1**, wherein the first caging groups inhibit the enzyme from acting upon the substrate by at least about 75%, at least about 90%, at least about 95%, or at least about 98%, as compared to the substrate in the absence of the first caging groups.

4. (Previously presented) The composition of claim **1**, wherein the first caging groups prevent the enzyme from acting upon the substrate.

5. (Previously presented) The composition of claim **1**, wherein removal of the first caging groups permits the enzyme to act upon the substrate.

6. (Previously presented) The composition of claim **1**, wherein the first label is an optically detectable label or a fluorophore; wherein the first signal and/or the second signal is an optical signal, a fluorescent signal, a luminescent signal, a nonoptical signal, or a magnetic signal; or wherein the first signal is a fluorescent emission at a first wavelength with a first intensity and the second signal is a fluorescent emission at the first wavelength with a second intensity substantially greater or less than the first intensity.

7. (Previously presented) The composition of claim **1**, wherein the one or more first caging groups associated with the one or more molecules are covalently attached to the one or more molecules.

8. (Previously presented) The composition of claim **1**, wherein the one or more first caging groups are removable by sonication, photoactivatable, or photolabile; or wherein the first caging groups can be removed by exposure to light with a wavelength between about 60 nm and about 400 nm, between about 400 nm and about 700 nm, and/or between about 700 nm and about 1000 nm.

9. (Previously presented) The composition of claim **1**, wherein the first label and the substrate are physically connected.

10. (Previously presented) The composition of claim **1**, wherein the substrate comprises one or more of: an amino acid, a polypeptide, a nitrogenous base, a nucleoside, a nucleotide, a nucleic acid, a carbohydrate, or a lipid.

11-12. (Cancelled)

13. (Original) The composition of claim **1**, wherein the enzyme is an oxidoreductase, a transferase, a hydrolase, a lyase, a ligase, an isomerase, a phosphatase, a GTPase, an ATPase, a phosphodiesterase, a luciferase, an acetylase, a glycosylase, a ubiquitin-conjugating enzyme, a hydrogenase, a polymerase, a peroxidase, a protease, or a caspase.

14. (Withdrawn) The composition of claim **13**, wherein the enzyme is a caspase, and wherein one polypeptide comprises the substrate for the caspase and the first label and

comprises a second label or a quencher; wherein the first label and the second label or the quencher interact to produce the first signal when the substrate is intact; and wherein cleavage of the substrate by the caspase prevents the interaction of the first label and the second label or the quencher, thereby resulting in production of the second signal.

15. (Withdrawn) The composition of claim **14**, wherein the caspase is caspase 3, the substrate comprises an Asp-Glu-Val-Asp motif, and the one or more first caging groups are located on one or more of the amino acid residues in the Asp-Glu-Val-Asp motif.

16. (Withdrawn) The composition of claim **14**, wherein one of the first label and the second label or the quencher is located at the N-terminus of the polypeptide and the other of the first label and the second label or the quencher is located at the C-terminus of the polypeptide.

17. (Withdrawn) The composition of claim **14**, wherein the first and second labels are fluorophores capable of exhibiting FRET, or wherein the first label is FITC and the second label is rhodamine or coumarin.

18. (Previously presented) The composition of claim **1**, wherein the enzyme is a protein kinase that phosphorylates tyrosine, serine and/or threonine.

19. (Previously presented) The composition of claim **18**, wherein the one or more molecules comprise one polypeptide; wherein the one polypeptide comprises the first label and the substrate for the kinase, the substrate comprising a serine, threonine, or tyrosine residue capable of being phosphorylated by the kinase; wherein the first label is located at the serine, threonine, or tyrosine residue and exhibits the first signal when the residue is not phosphorylated and the second signal when the residue is phosphorylated.

20. (Cancelled)

21. (Previously presented) A composition, comprising:

a cell comprising an enzyme and a caged sensor for detecting an activity of the enzyme, which caged sensor comprises:

a) one or more molecules collectively comprising:

i) a substrate for the enzyme, wherein the substrate is in a first state on which the enzyme can act, thereby converting the substrate to a second state, and

ii) a first label, wherein a first signal exhibited by the first label when the substrate is in its first state is distinguishable from a second signal exhibited by the first label when the substrate is in its second state; and,

b) one or more first caging groups associated with the one or more molecules, the first caging groups inhibiting the enzyme from acting upon the substrate;

wherein the enzyme is a protein kinase that phosphorylates tyrosine, serine and/or threonine;

wherein the one or more molecules comprise one polypeptide; wherein the one polypeptide comprises the substrate for the kinase and the first label and comprises a second label or a quencher; wherein the first label and the second label or the quencher interact to produce the first signal when the substrate is not phosphorylated; and wherein phosphorylation of the substrate prevents the interaction of the first label and the second label or the quencher, thereby resulting in production of the second signal.

22. (Previously presented) The composition of claim **21** or **304**, wherein the one or more first caging groups are located on a residue that can be phosphorylated by the kinase.

23. (Previously presented) The composition of claim **21** or **304**, wherein one of the first label and the second label or the quencher is located at the N-terminus of the polypeptide and the other of the first label and the second label or the quencher is located at the C-terminus of the polypeptide.

24. (Previously presented) The composition of claim **21** or **304**, wherein the first and second labels are hydrophobic fluorophores, or wherein the first label is FITC and the second label is rhodamine or coumarin.

25. (Previously presented) The composition of claim **21** or **304**, wherein phosphorylation of the substrate triggers a conformational change in the polypeptide, the conformational change preventing the interaction of the first label and the second label or the quencher; or wherein phosphorylation of the substrate results in binding of a phosphobinder to the phosphorylated

substrate, the binding of the phosphobinder preventing the interaction of the first label and the second label or the quencher.

26. (Original) The composition of claim **25**, wherein the phosphobinder is associated with one or more second caging groups, the presence of which prevents the phosphobinder from binding the phosphorylated substrate.

27. (Original) The composition of claim **26**, wherein the second caging groups are removable under different conditions than the first caging groups preventing phosphorylation of the substrate.

28. (Original) The composition of claim **25**, wherein the phosphobinder comprises an antibody, an SH-2 domain, a PTB domain, a 14-3-3 domain, an FHA domain, a WD40 domain and/or a WW domain.

29. (Previously presented) A composition, comprising:

a cell comprising an enzyme and a caged sensor for detecting an activity of the enzyme, which caged sensor comprises:

a) one or more molecules collectively comprising:

i) a substrate for the enzyme, wherein the substrate is in a first state on which the enzyme can act, thereby converting the substrate to a second state, and

ii) a first label, wherein a first signal exhibited by the first label when the substrate is in its first state is distinguishable from a second signal exhibited by the first label when the substrate is in its second state; and,

b) one or more first caging groups associated with the one or more molecules, the first caging groups inhibiting the enzyme from acting upon the substrate;

wherein the enzyme is a protein kinase that phosphorylates tyrosine, serine and/or threonine;

wherein the one or more molecules comprise one polypeptide; wherein the one polypeptide comprises the substrate for the kinase and the first label; wherein the polypeptide comprises a phosphobinder and a second label or a quencher; wherein the first label and the second label or the quencher do not interact when the substrate is not phosphorylated,

thereby producing the first signal; and wherein phosphorylation of the substrate results in intramolecular binding of the phosphobinder to the phosphorylated substrate, the intramolecular binding resulting in the interaction of the first label and the second label or the quencher, thereby producing the second signal; or, wherein the one or more molecules comprise a first polypeptide and a second polypeptide; wherein the first polypeptide comprises the substrate for the kinase and the first label; wherein the second polypeptide comprises a phosphobinder and a second label or a quencher; wherein the first label and the second label or the quencher do not interact when the substrate is not phosphorylated, thereby producing the first signal; and wherein phosphorylation of the substrate results in intermolecular binding of the phosphobinder to the phosphorylated substrate, the intermolecular binding resulting in the interaction of the first label and the second label or the quencher, thereby producing the second signal.

30. (Previously presented) The composition of claim **29** or **305**, wherein the one or more first caging groups are located on a residue that can be phosphorylated by the kinase.

31. (Previously presented) The composition of claim **29** or **305**, wherein one of the first label and the second label or the quencher is located at the N-terminus of the polypeptide and the other of the first label and the second label or the quencher is located at the C-terminus of the polypeptide.

32. (Previously presented) The composition of claim **29** or **305**, wherein the first and second labels are fluorophores capable of exhibiting FRET.

33. (Previously presented) The composition of claim **29** or **305**, wherein the phosphobinder is associated with one or more second caging groups, the presence of which prevents the phosphobinder from binding the phosphorylated substrate.

34. (Original) The composition of claim **33**, wherein the second caging groups are removable under different conditions than the first caging groups preventing phosphorylation of the substrate.

35. (Previously presented) The composition of claim **29** or **305**, wherein the phosphobinder comprises an antibody, an SH-2 domain, a PTB domain, a 14-3-3 domain, an FHA domain, a WD40 domain and/or a WW domain.

36. (Previously presented) A composition, comprising:

a cell comprising an enzyme and a caged sensor for detecting an activity of the enzyme, which caged sensor comprises:

a) one or more molecules collectively comprising:

i) a substrate for the enzyme, wherein the substrate is in a first state on which the enzyme can act, thereby converting the substrate to a second state, and

ii) a first label, wherein a first signal exhibited by the first label when the substrate is in its first state is distinguishable from a second signal exhibited by the first label when the substrate is in its second state; and,

b) one or more first caging groups associated with the one or more molecules, the first caging groups inhibiting the enzyme from acting upon the substrate;

wherein the enzyme is a protein kinase that phosphorylates tyrosine, serine and/or threonine;

wherein the one or more molecules comprise one polypeptide; wherein the one polypeptide comprises the substrate for the kinase, a second substrate, the first label, a third label, a fourth label or a quencher, and a phosphobinder; the substrate comprising a serine, threonine, or tyrosine residue capable of being phosphorylated by the kinase; the second substrate being associated with one or more third caging groups, the presence of which prevents phosphorylation of the second substrate; wherein the first label is located at the serine, threonine, or tyrosine residue and exhibits the first signal when the residue is not phosphorylated and the second signal when the residue is phosphorylated; wherein the third label and the fourth label or the quencher do not interact when the second substrate is not phosphorylated, thereby producing a third signal; and wherein phosphorylation of the second substrate results in intramolecular binding of the phosphobinder to the phosphorylated second substrate, the intramolecular binding resulting in the interaction of the third label and the fourth label or the quencher, thereby producing a fourth signal, the fourth signal distinguishable from the first, second and third signals.

37. (Previously presented) The composition of claim **36** or **306**, wherein the second substrate is for the same kinase or for a different kinase.

38. (Previously presented) The composition of claim **36** or **306**, wherein the one or more third caging groups are located on a residue that can be phosphorylated by the kinase.

39. (Original) The composition of claim **38**, wherein the third caging groups preventing phosphorylation of the second substrate are removable under different conditions than the first caging groups preventing phosphorylation of the substrate.

40. (Previously presented) The composition of claim **36** or **306**, wherein one of the third label and the fourth label or the quencher is located at the C-terminus of the polypeptide and the other of the third label and the fourth label or the quencher is within the polypeptide.

41. (Previously presented) The composition of claim **36** or **306**, wherein the third and fourth labels are fluorophores capable of exhibiting FRET.

42. (Previously presented) The composition of claim **36** or **306**, wherein the phosphobinder is associated with one or more second caging groups, the presence of which prevents the phosphobinder from binding the phosphorylated second substrate.

43. (Original) The composition of claim **42**, wherein the second caging groups are removable under different conditions than the first caging groups preventing phosphorylation of the substrate and/or under different conditions than the third caging groups preventing phosphorylation of the second substrate.

44. (Previously presented) The composition of claim **36** or **306**, wherein the phosphobinder comprises an antibody, an SH-2 domain, a PTB domain, a 14-3-3 domain, an FHA domain, a WD40 domain and/or a WW domain.

45. (Previously presented) A composition, comprising:

a cell comprising an enzyme and a caged sensor for detecting an activity of the enzyme, which caged sensor comprises:

a) one or more molecules collectively comprising:

i) a substrate for the enzyme, wherein the substrate is in a first state on which the enzyme can act, thereby converting the substrate to a second state, and

ii) a first label, wherein a first signal exhibited by the first label when the substrate is in its first state is distinguishable from a second signal exhibited by the first label when the substrate is in its second state, and,

b) one or more first caging groups associated with the one or more molecules, the first caging groups inhibiting the enzyme from acting upon the substrate;

wherein the one or more molecules comprise a fifth label, the fifth label exhibiting a unique fifth signal, the fifth signal being independent of the state of the substrate.

46. (Previously presented) The composition of claim **45** or **307**, wherein the fifth label is a fluorophore or a quantum dot.

47. (Previously presented) The composition of claim **1**, wherein the one or more molecules are associated with a cellular delivery module that can mediate introduction of the sensor into a cell.

48. (Original) The composition of claim **47**, wherein the cellular delivery module comprises a polypeptide, a PEP-1 peptide, an amphipathic peptide, a cationic peptide, a protein transduction domain, a protein transduction domain derived from an HIV-1 Tat protein, from a herpes simplex virus VP22 protein, or from a Drosophila antennapedia protein, or a model protein transduction domain.

49. (Original) The composition of claim **47**, wherein the cellular delivery module is covalently attached to the one or more molecules.

50. (Original) The composition of claim **49**, wherein the covalent attachment is reversible by exposure to light of a preselected wavelength.

51. (Original) The composition of claim **47**, wherein the cellular delivery module is associated with one or more fourth caging groups, the presence of which prevents the cellular delivery module from mediating introduction of the sensor into a cell.

52. (Previously presented) The composition of claim **1**, wherein the one or more molecules are associated with at least one subcellular delivery module.

53. (Original) The composition of claim **52**, wherein the subcellular delivery module comprises a polypeptide, a nucleic acid, and/or a carbohydrate; wherein the subcellular

delivery module mediates localization of the sensor to one or more of: a membrane, a mitochondrion, a peroxisome, a nucleus, an endoplasmic reticulum, a Golgi, a vesicle, a lysosome, an endosome, or a chloroplast; wherein the subcellular delivery module comprises one or more of: a mitochondrial matrix-targeting sequence, a nuclear localization signal, a signal peptide, an ER retention signal, a peroxisomal targeting motif, a chloroplast stromal targeting sequence, a transmembrane domain, or a lipid attachment site; or wherein the subcellular delivery module comprises a binding domain that mediates localization of the sensor by binding to a target protein.

54. (Original) The composition of claim **52**, wherein the subcellular delivery module is covalently attached to the one or more molecules.

55. (Original) The composition of claim **54**, wherein the covalent attachment is reversible by exposure to light of a preselected wavelength.

56. (Original) The composition of claim **52**, wherein the subcellular delivery module is associated with one or more fifth caging groups, the presence of which prevents the subcellular delivery module from mediating subcellular localization of the sensor.

57-60. (Cancelled)

61. (Previously presented) A kit for making the caged sensor of claim **1**, comprising a substrate, a first label, one or more first caging groups, and instructions for assembling the substrate, the first label, and the first caging groups to form the caged sensor, packaged in one or more containers; or comprising a first label, one or more first caging groups, and instructions for assembling the first label, the first caging groups, and a substrate supplied by a user of the kit to form the caged sensor, packaged in one or more containers.

62-303. (Cancelled)

304. (Previously presented) A composition, comprising:

an enzyme and a caged sensor for detecting an activity of the enzyme, which caged sensor comprises:

a) one or more molecules collectively comprising:

i) a substrate for the enzyme, wherein the substrate is in a first state on which the enzyme can act, thereby converting the substrate to a second state, wherein the first state is not converted to the second state by cleavage by the enzyme, and

ii) a first label, wherein a first signal exhibited by the first label when the substrate is in its first state is distinguishable from a second signal exhibited by the first label when the substrate is in its second state; and,

b) one or more first caging groups associated with the one or more molecules, the first caging groups inhibiting the enzyme from acting upon the substrate;

wherein the enzyme is a protein kinase that phosphorylates tyrosine, serine and/or threonine;

wherein the one or more molecules comprise one polypeptide; wherein the one polypeptide comprises the substrate for the kinase and the first label and comprises a second label or a quencher; wherein the first label and the second label or the quencher interact to produce the first signal when the substrate is not phosphorylated; and wherein phosphorylation of the substrate prevents the interaction of the first label and the second label or the quencher, thereby resulting in production of the second signal.

305. (Previously presented) A composition, comprising:

an enzyme and a caged sensor for detecting an activity of the enzyme, which caged sensor comprises:

a) one or more molecules collectively comprising:

i) a substrate for the enzyme, wherein the substrate is in a first state on which the enzyme can act, thereby converting the substrate to a second state, wherein the first state is not converted to the second state by cleavage by the enzyme, and

ii) a first label, wherein a first signal exhibited by the first label when the substrate is in its first state is distinguishable from a second signal exhibited by the first label when the substrate is in its second state; and,

b) one or more first caging groups associated with the one or more molecules, the first caging groups inhibiting the enzyme from acting upon the substrate;

wherein the enzyme is a protein kinase that phosphorylates tyrosine, serine and/or threonine;

wherein the one or more molecules comprise one polypeptide; wherein the one polypeptide comprises the substrate for the kinase and the first label; wherein the polypeptide comprises a phosphobinder and a second label or a quencher; wherein the first label and the second label or the quencher do not interact when the substrate is not phosphorylated, thereby producing the first signal; and wherein phosphorylation of the substrate results in intramolecular binding of the phosphobinder to the phosphorylated substrate, the intramolecular binding resulting in the interaction of the first label and the second label or the quencher, thereby producing the second signal; or, wherein the one or more molecules comprise a first polypeptide and a second polypeptide; wherein the first polypeptide comprises the substrate for the kinase and the first label; wherein the second polypeptide comprises a phosphobinder and a second label or a quencher; wherein the first label and the second label or the quencher do not interact when the substrate is not phosphorylated, thereby producing the first signal; and wherein phosphorylation of the substrate results in intermolecular binding of the phosphobinder to the phosphorylated substrate, the intermolecular binding resulting in the interaction of the first label and the second label or the quencher, thereby producing the second signal.

306. (Previously presented) A composition, comprising:

an enzyme and a caged sensor for detecting an activity of the enzyme, which caged sensor comprises:

a) one or more molecules collectively comprising:

i) a substrate for the enzyme, wherein the substrate is in a first state on which the enzyme can act, thereby converting the substrate to a second state, wherein the first state is not converted to the second state by cleavage by the enzyme, and

ii) a first label, wherein a first signal exhibited by the first label when the substrate is in its first state is distinguishable from a second signal exhibited by the first label when the substrate is in its second state; and,

b) one or more first caging groups associated with the one or more molecules, the first caging groups inhibiting the enzyme from acting upon the substrate;

wherein the enzyme is a protein kinase that phosphorylates tyrosine, serine and/or threonine;

wherein the one or more molecules comprise one polypeptide; wherein the one polypeptide comprises the substrate for the kinase, a second substrate, the first label, a third label, a fourth label or a quencher, and a phosphobinder; the substrate comprising a serine, threonine, or tyrosine residue capable of being phosphorylated by the kinase; the second substrate being associated with one or more third caging groups, the presence of which prevents phosphorylation of the second substrate; wherein the first label is located at the serine, threonine, or tyrosine residue and exhibits the first signal when the residue is not phosphorylated and the second signal when the residue is phosphorylated; wherein the third label and the fourth label or the quencher do not interact when the second substrate is not phosphorylated, thereby producing a third signal; and wherein phosphorylation of the second substrate results in intramolecular binding of the phosphobinder to the phosphorylated second substrate, the intramolecular binding resulting in the interaction of the third label and the fourth label or the quencher, thereby producing a fourth signal, the fourth signal distinguishable from the first, second and third signals.

307. (Previously presented) A composition, comprising:

an enzyme and a caged sensor for detecting an activity of the enzyme, which caged sensor comprises:

a) one or more molecules collectively comprising:

i) a substrate for the enzyme, wherein the substrate is in a first state on which the enzyme can act, thereby converting the substrate to a second state, wherein the first state is not converted to the second state by cleavage by the enzyme, and

ii) a first label, wherein a first signal exhibited by the first label when the substrate is in its first state is distinguishable from a second signal exhibited by the first label when the substrate is in its second state, and,

b) one or more first caging groups associated with the one or more molecules, the first caging groups inhibiting the enzyme from acting upon the substrate;

wherein the one or more molecules comprise a fifth label, the fifth label exhibiting a unique fifth signal, the fifth signal being independent of the state of the substrate.

308. (Previously presented) The composition of claim **1**, wherein an induced conformational change in the first caging groups permits the enzyme to act upon the substrate.